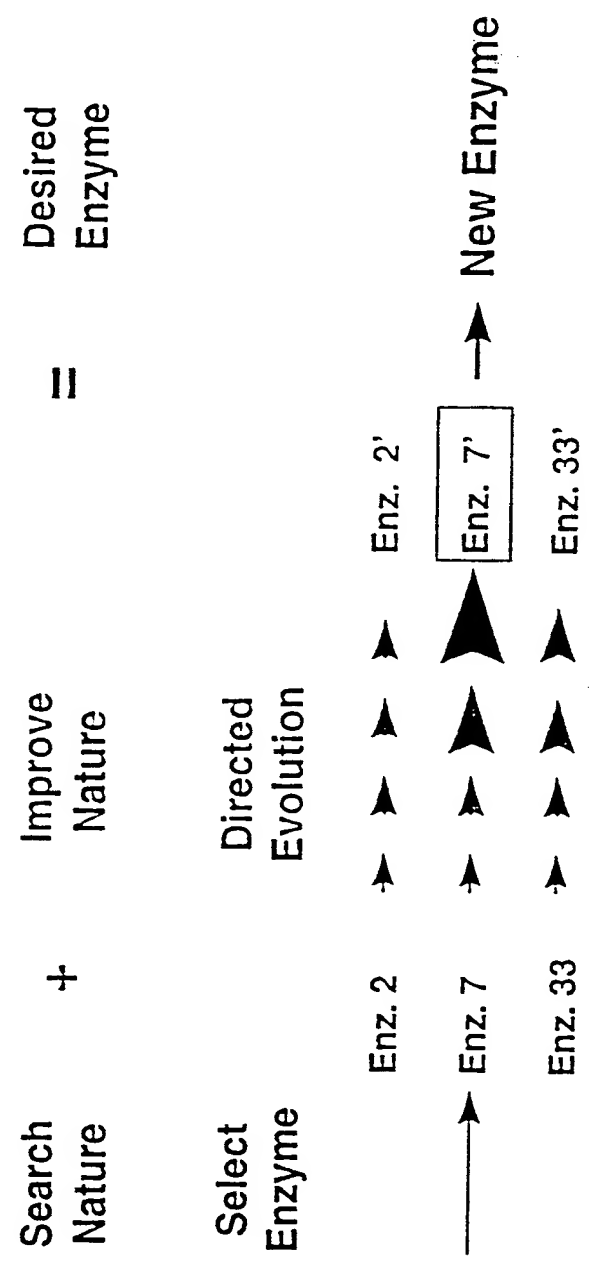


NA Library

Combinatorial Enzyme Development

(Natural + Non-natural Evolution)



ID via High Throughput Screening

ID via Enzyme Characterization ID via Mutation / Selection

Figure 12

Bypassing Barriers to Directed Protein Evolution

(Barrier = Capacity limit of directed evolution system)

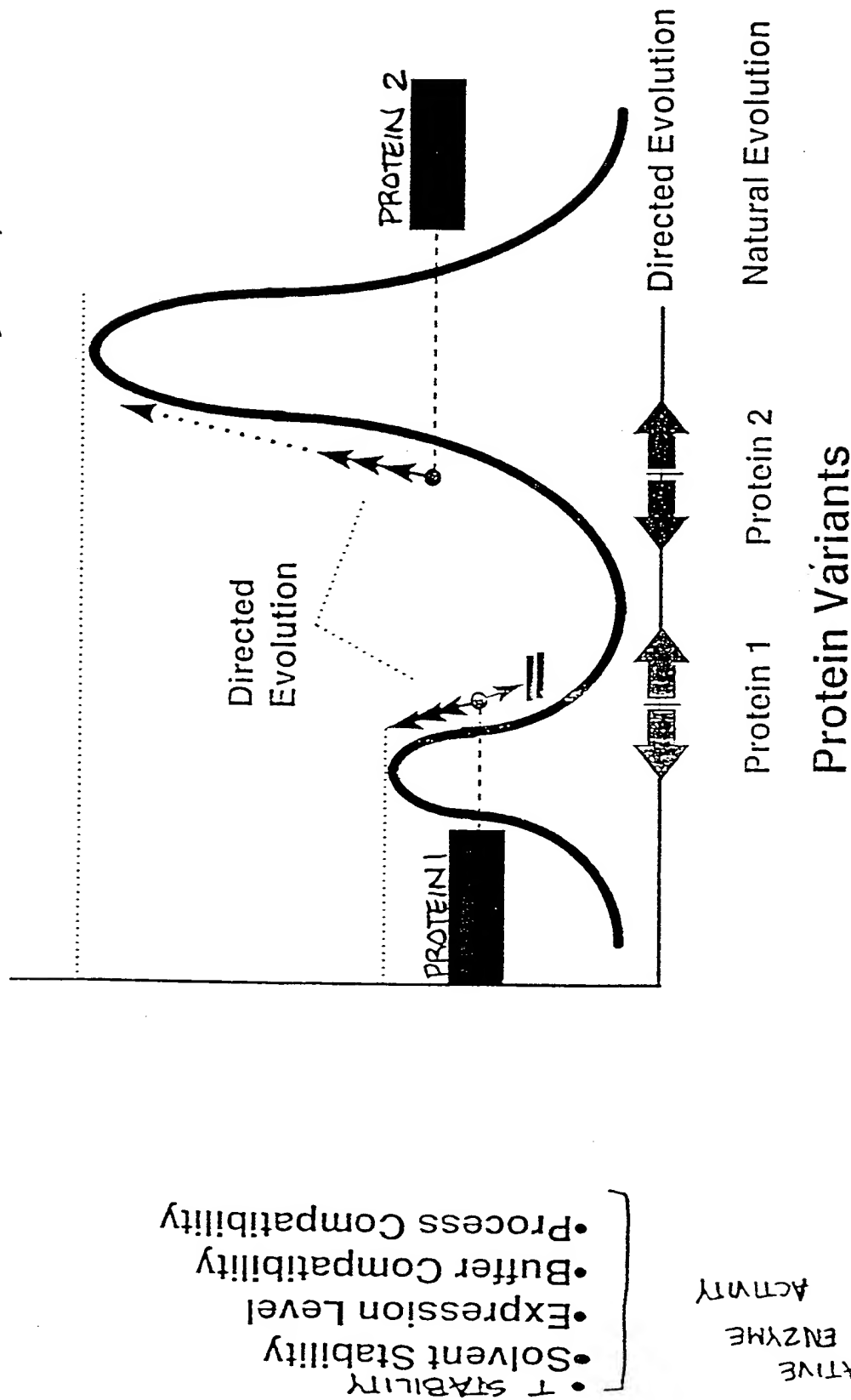
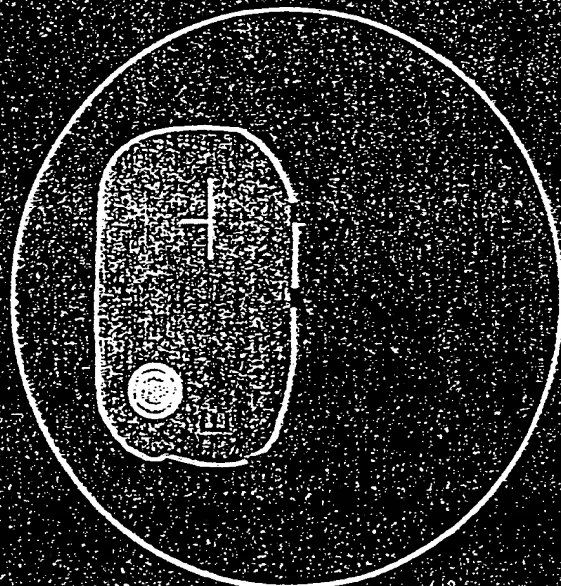


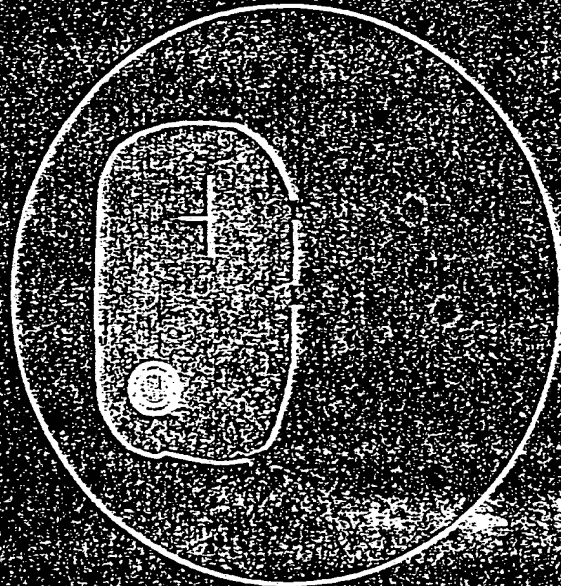
Figure 13



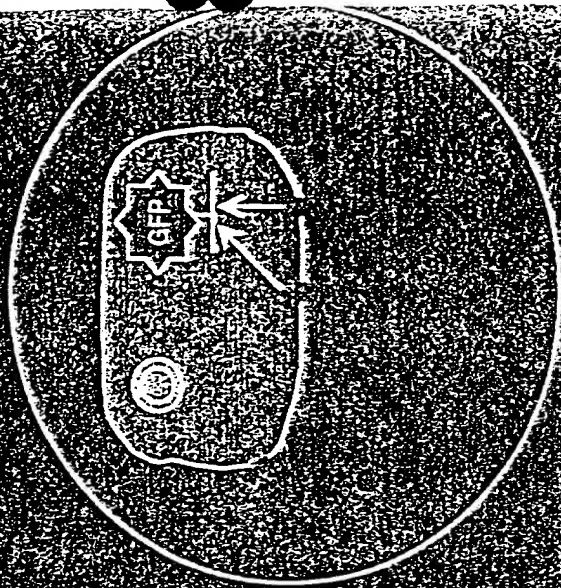
Co-Encapsulation Assay for Novel Bioactive Discovery



Co-encapsulation
Library + Eukaryote



Growth and expression
of small molecule from host



Receptor binding of small
molecule & GFP reporting

E = Eukaryotic assay organism L = Large Insert library SM = Small molecule
GFP = Green Fluorescent Protein R = Eukaryotic receptor



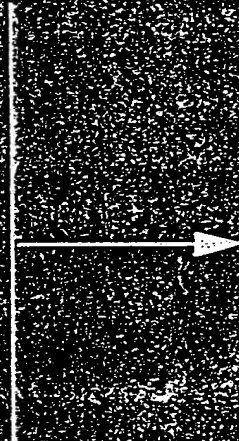
AMERICAN

FACS Screening for Encapsulation

Test organisms

Pathway clones

encapsulate



live/dead or other
activity stain



sort

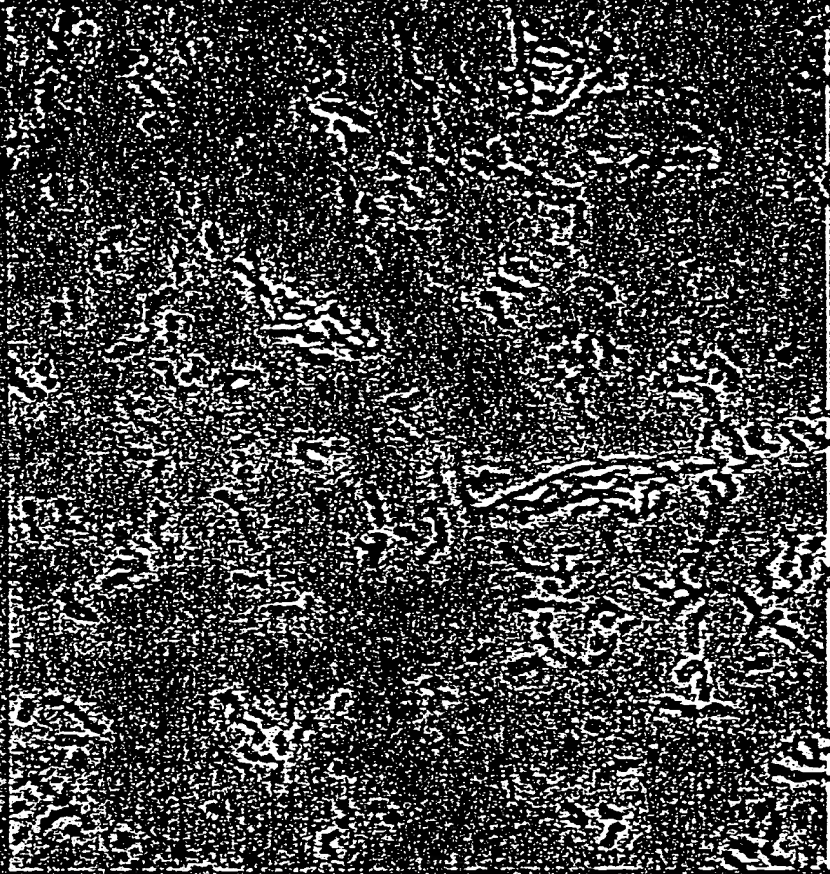
bioactive expression

(e.g. live/dead, growth rate,
metabolic stains etc.)

Figure 15



Development of Unicellular *Streptomyces* Strains for Ultra-High Throughput Screening



Streptomyces lividans
mycelia

Streptomyces "diversa"
unicells



FACS Enrichment for Gel Microdroplets Containing *S. diversa*

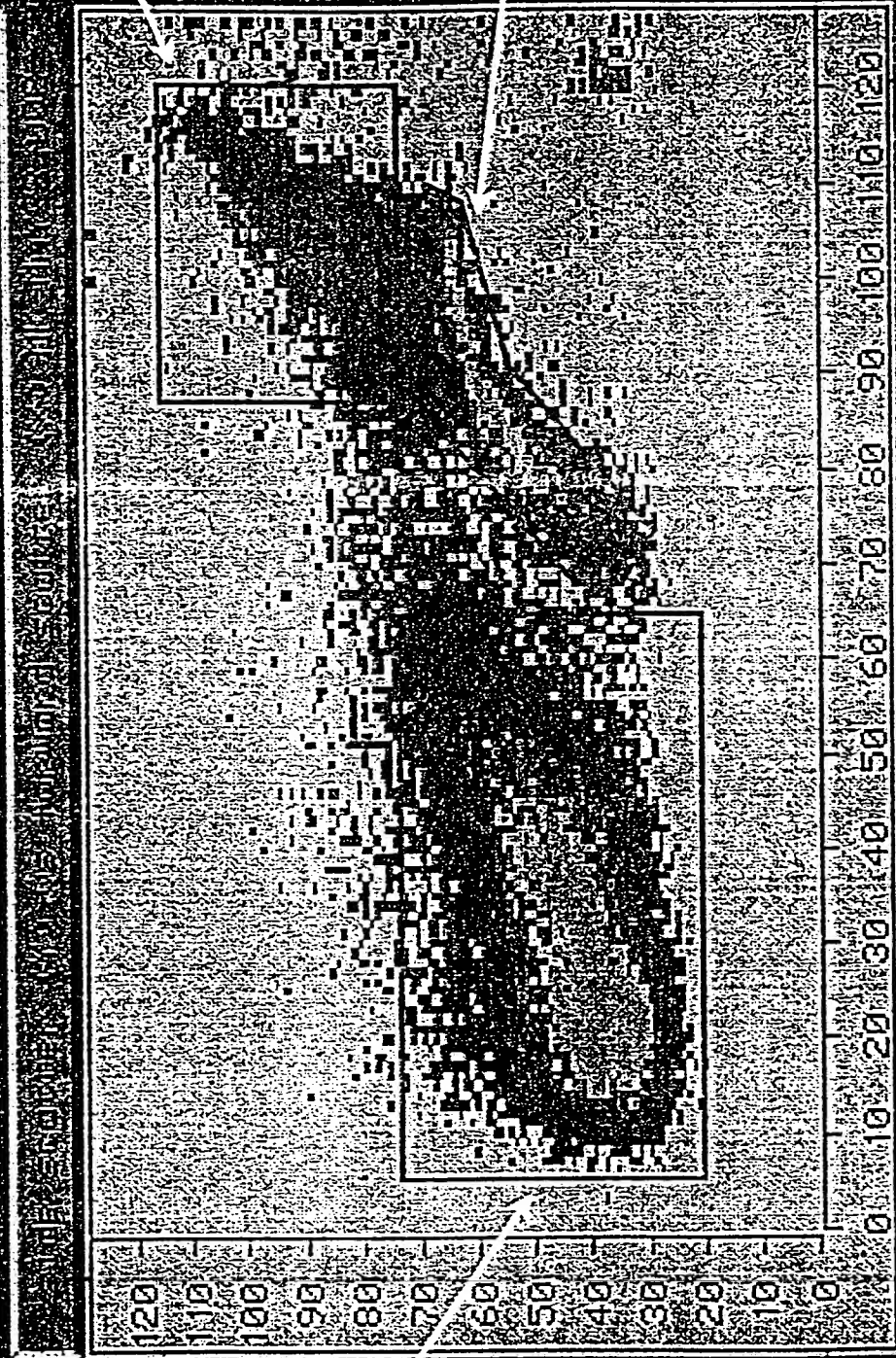


Figure 17